S-CHOLESTERYL ALKANETHIOATES

Wolfgang Elser, et al

Army Electronics Command
Fort Belvoir, Virginia

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WOLFGANG ELSER, JUERGEN L. W. POHLMANN, and PHILLIP R. BOYD

U.S. Army Electronics Command
Night Vision Laboratory
Ft. Belvoir, Va. 22060

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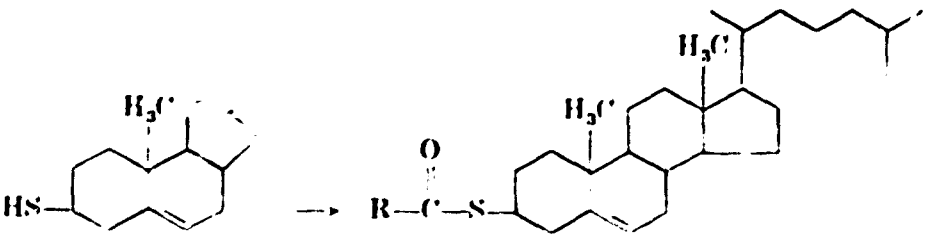
Abstract A homologous series of S-cholesteryl alkanethioates was prepared by reaction of 3β-mercaptocholest-5-ene with the imidazolides of alkanoic acids. The mesomorphic properties of 20 members, S-cholesteryl thioformate through eicosanethioate were determined. All alkanethioates are monotropic cholesteric except the cholesteryl thioformate. Cholesteryl thioheptanoate and all higher homologues in this series also exhibit a monotropic smectic mesophase. A comparison with the corresponding cholesteryl alkanoates shows that the replacement of the ester group by the thiol ester group leads to an increase in both the smectic-cholesteric and the cholesteric-isotropic transition temperatures.

In earlier communications, we described the mesomorphic behavior of 3β-steryl alkyl carbonates[1,2] and thiocarbonates[3,4] and established that the thiocarbonate group gives an increase in transition temperatures and a decrease in transition heats[3] as compared with the carbonate group. We also reported briefly that similar relations were found between the ester and the thiol ester group.[5] This paper is concerned with a homologous series of S-cholesteryl alkanethioates and their mesomorphic behavior.

Cholesteryl alkanoates generally are obtained by esterification of cholesterol with free fatty acids,[6,7] acid anhydrides,[8] or acid chlorides.[8,9] These established methods usually give poor to moderate yields and often present considerable difficulties in the purification of the cholesteryl esters by recrystallization. Two other reactions have recently been reported. A transesterification procedure, followed by chromatographic purification, gives cholesteryl esters in good yield and a high state of purity, but is useful only for cholesteryl esters of higher fatty acids.[10] The other method is the transacylation of imidazolides.[11] The reaction of carboxylic acids

with 1,1′-carbonyldiimidazole combined with alcoholysis leads to esters under particularly mild reaction conditions.[12] The latter was successfully used in the preparation of cholesteryl esters of hydroxy, keto, and epoxy fatty acids,[13] and also gave excellent yields in the preparation of cholesteryl alkanoates[14] and other 3β-steryl esters.[15] Because of the mild reaction conditions, this method was chosen to synthesize the fatty acid esters of 3β-mercaptocholest-5-ene.

The use of 1,1′-carbonyldiimidazole in the synthesis of S-cholesteryl alkanethioates (2) from 3β-mercaptocholest-5-ene (1) and alkanoic acids in absolute benzene gave excellent yields for the lower members, but generally somewhat lower yields for the higher members (Table 1). The use of a solvent of higher polarity did not substantially increase the yield for the higher members.



1 2

The abnormally low yield of S-cholesteryl thioformate is explained by the fact that 1-formylimidazole decomposes almost quantitatively into carbon monoxide and imidazole above its melting point,[16] a concurrent reaction to the formation of the thioformate. No attempt was made to increase the yield by catalysis with sodium ethoxide or imidazolylsodium.[17,18]

Only two contaminants, unreacted 3β-mercaptocholest-5-ene and dicholesteryl disulfide,[19] were found in the reaction products of this transacylation. Both were identified by thin-layer chromatography (Table 2) and could be removed by column chromatographic purification. 3β-Mercaptocholest-5-ene as a thiol oxidizes[20] easily, and therefore traces of dicholesteryl disulfide are always found in commercial thiocholesterol. However, after recrystallization from ethyl acetate, it showed the reported physical constants[19,20,21] and was also free of cholesterol (Table 2).

The transition curves of the homologous series of S-cholesteryl

TABLE I S-Cholesteryl Alkanethioates

Alkanethioate	Yield (%)	m.p. (°C)	S-Ch[a]	Ch-I[b]	Formula	Mol. wt.	Analytical values (%) Calculated C	H	S	Found C	H	S
Thioformate	31[c]	118.3			$C_{28}H_{46}OS$	431.7	77.90	10.97	7.43	77.93	10.89	7.68
Thioacetate	85	126.2[d]		119.5	$C_{29}H_{48}OS$	445.8	78.14	11.08	7.19	78.17	10.98	7.23
Thiopropionate	65	111.4		111.1	$C_{30}H_{50}OS$	459.8	78.37	11.18	6.97	78.65	11.09	7.09
Thiobutyrate	78	100.6		117.6	$C_{31}H_{52}OS$	473.8	78.58	11.27	6.77	78.34	11.11	6.95
Pentanethioate	64	90.7		104.7	$C_{32}H_{54}OS$	487.8	78.78	11.36	6.57	78.88	11.25	6.84
Hexanethioate	66	94.8		107.7	$C_{33}H_{56}OS$	501.9	78.98	11.45	6.39	79.22	11.37	6.35
Heptanethioate	77	107.0	73.1	102.1	$C_{34}H_{58}OS$	515.9	79.16	11.53	6.21	79.30	11.33	6.28
Octanethioate	93	97.3	76.6	100.8	$C_{35}H_{60}OS$	529.9	79.33	11.60	6.05	79.24	11.65	6.11
Nonanethioate	83	68.2	84.0	97.5	$C_{36}H_{62}OS$	544.0	79.49	11.67	5.89	79.64	11.50	5.82
Decanethioate	91	79.5	87.4	98.3	$C_{37}H_{64}OS$	558.0	79.64	11.74	5.75	78.89	11.90	5.96
Undecanethioate	72	80.6	88.1	95.0	$C_{38}H_{66}OS$	572.0	79.79	11.81	5.60	80.01	11.57	5.78
Dodecanethioate	67	83.5	86.7	92.3	$C_{39}H_{68}OS$	586.0	79.93	11.87	5.47	80.10	11.75	5.70
Tridecanethioate	61	78.9	86.5	91.4	$C_{40}H_{70}OS$	600.1	80.06	11.93	5.34	80.19	11.86	5.50
Tetradecanethioate	62	72.2	85.5	90.4	$C_{41}H_{72}OS$	614.1	80.19	11.98	5.22	80.07	11.83	5.48
Pentadecanethioate	76	61.3	84.3	88.3	$C_{42}H_{74}OS$	628.1	80.31	12.04	5.10	80.13	12.05	5.33
Hexadecanethioate	74	57.0	83.0	87.3	$C_{43}H_{76}OS$	642.2	80.43	12.09	4.99	80.54	12.07	5.26
Heptadecanethioate	66	67.5	81.2	85.0	$C_{44}H_{78}OS$	656.2	80.54	12.14	4.88	80.47	12.17	5.09
Octadecanethioate	74	64.5	80.2	84.2	$C_{45}H_{80}OS$	670.2	80.65	12.18	4.78	80.81	12.31	5.04
Nonadecanethioate	66	73.5	78.3	82.4	$C_{46}H_{82}OS$	684.2	80.75	12.23	4.68	80.96	12.41	4.90
Eicosanethioate	60	71.2	77.1	81.3	$C_{47}H_{84}OS$	698.3	80.85	12.27	4.59	81.06	12.22	4.88

(a) smectic-cholesteric transition, °C.

(b) cholesteric-isotropic transition, °C.

(c) no attempt was made to increase yield by different technique.

(d) S. Bernstein and K. J. Sax[30] report m.p. 122.5-124.5°.

TABLE 2 R_F-values on Silica Gel (Merck), Precoated Plates

Solvent: Benzene-hexane 15:85 (v/v)

cholesterol	0.08
3β-mercaptocholest-5-ene	0.62
dicholesteryl disulfide	0.34
S-cholesteryl thioacetate	0.21
S-cholesteryl octanethioate	0.40
S-cholesteryl tetradecanethioate	0.45
S-cholesteryl eicosanethioate	0.48

alkanethioates are shown in Fig. 1. Monotropic smectic mesophases are exhibited by S-cholesteryl heptanethioate and all higher members of the series. Monotropic cholesteric mesophases are found in all members except the thioformate. Cholesteric colors are exhibited

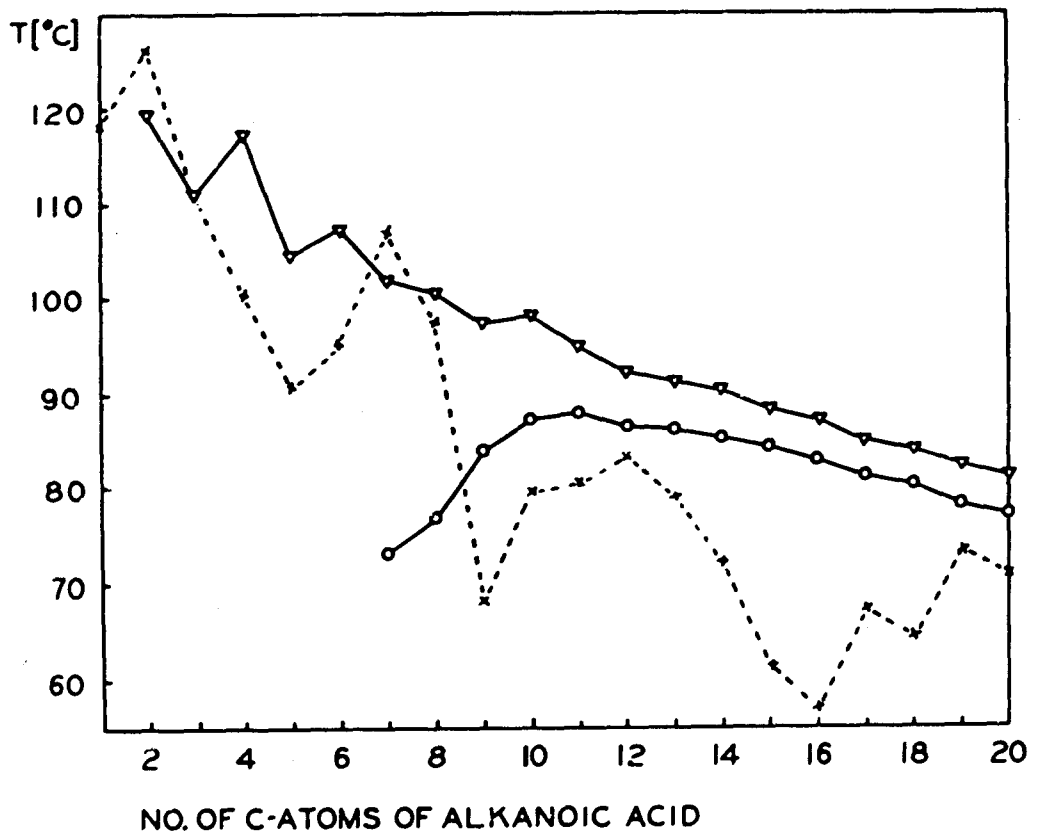


Figure 1. Transition temperatures of S-cholesteryl alkanethioates: X, melting points; ▽, ○, cholesteric-isotropic transitions; ▼, smectic-cholesteric transitions.

only by S-cholesteryl pentanethioate, hexanethioate, heptanethioate, and octanethioate; the eicosanethioate shows "crystal colors",[3] *i.e.*, regions of intense colors co-existing with crystals over a wide temperature range.

Comparing these data with those of the corresponding cholesteryl alkanoates[5] shows that replacing the ester group by the thiol ester group results in an increase of both the smectic-cholesteric and the cholesteric-isotropic transition temperatures. In addition, it leads to the existence of a smectic mesophase in the S-cholesteryl heptanethioate and the octanethioate, not observed in the corresponding cholesteryl alkanoates. The cholesteric-isotropic transition points of the first half of the series show an odd-even effect and thereafter a steady decrease with chain length. The smectic-cholesteric transition curve first increases and then decreases. This results in a narrowing of the range of the cholesteric mesophase until an almost constant value is reached with the dodecanethioate. This behavior seems to be typical for homologous series exhibiting both smectic and cholesteric mesophases.[3,4]

Experimental

STARTING MATERIALS. Commercial 3β-mercaptocholest-5-ene[22] was used after two recrystallizations from ethyl acetate, mp 97–97.5°, and 1,1′-carbonyldiimidazole[22] was employed as received. The alkanoic acids used had a minimum purity of 99%, with the exception of pentadecanoic acid which was only 98% pure.

PREPARATION OF MATERIALS. As a typical example, the preparation of S-cholesteryl tetradecanethioate is given. Monitoring the reaction by thin-layer chromatography revealed that after 2 hr an appreciable amount of 3β-mercaptocholest-5-ene was still present. Therefore, the esterification period was extended to 6 hr, whereafter no increase in yield was obtainable. Reaction times and molar ratios of reactants were the same. All experiments were carried out under nitrogen.

S CHOLESTERYL TETRADECANETHIOATE. A solution of 2.28 g (0.01 mol) of tetradecanoic acid in 10 ml of absolute benzene was added to a stirred slurry of 1.62 g (0.01 mol) of 1,1′-carbonyldiimidazole in 25 ml of absolute benzene. After about 20 min all material was dissolved and evolution of CO_2 had ceased. 4.03 g (0.01 mol)

of 3β-mercaptocholest-5-ene was added and the mixture was refluxed. After 6 hr the bulk of the benzene was distilled off, 100 ml of hexane added, the precipitated imidazole filtered off, and the filtrate chromatographed on a 45 × 350-mm column of silica gel.[23] Elution was performed with benzene-hexane 15:85 (v/v) and 100-ml fractions were collected. Thin-layer chromatography showed the tetradecanethioate in fractions 12–19, which were combined and evaporated to dryness. The residue was recrystallized twice from ethanol-butanone. Yield: 3.8 g (62%) of fine needles, mp 71 °; cp 89 °.

The physical properties of the prepared S-cholesteryl alkanethioates and their elemental analyses[24] are summarized in Table 1. The yields are expressed in analytically pure material, *i.e.*, after column chromatography and recrystallization. Transition points were determined with a Mettler FP-11 hot stage and are corrected. Methods for identification of phases[25] and measurements of heats of transition have already been published by this laboratory.[5]

PURITY. Since traces of dicholesteryl disulfide do not interfere with the reaction and are removed by column chromatographic purification of the alkanethioates, commercial 3β-mercaptocholest-5-ene[22] was used throughout the experiments. To check for other contaminants not revealed by thin-layer chromatography under the conditions employed for the analysis of the alkanethioates, cholesterol was purified by the bromination-debromination method of Fieser.[26] Two-dimensional thin-layer chromatography did not reveal any impurities.[27] 3β-Mercaptocholest-5-ene prepared from this pure cholesterol with the intermediate isothiuronium tosylate isolated and recrystallized[20] was used to prepare S-cholesteryl hexanethioate and heptanethioate. A comparison with the corresponding S-cholesteryl alkanethioates prepared from commercial 3β-mercaptocholest-5-ene did not reveal any discrepancies in the transition points of both pure and mixed samples.

The purity of the alkanoic acids used was determined by gas-liquid chromatography of their methyl esters[28] on an EGSS-X phase.[14] They were found to be 99% to 99.5%[29] pure. The only exception was pentadecanoic acid with a purity of 98%. The identified impurities were in most cases adjacent homologues. Unsaturated fatty acids, if present at all, were less than 0.1%.

Because of the known purity of the starting materials and the

complete removal of unreacted 3β-mercaptocholest-5-ene and dicholesteryl disulfide by column chromatography on silica gel, the homologous series of S-cholesteryl alkanethioates can be considered to have a minimum purity of 99%, with the possible exception of the pentadecanethioate, which might be only 98% pure.

MESOMORPHIC BEHAVIOR. The mesophases were determined by optical means[30] and temperatures of phase transitions with a microscopic hot stage (Mettler FP-11) and by differential scanning calorimetry (see data in Fig. 1). Since the transition heats in the melt have already appeared in another context,[5] only observations in the capillary[31] of compounds exhibiting cholesteric colors are listed.

S-CHOLESTERYL PENTANETHIOATE melts at 91–92° and clears at 102°. On cooling, the visible spectrum is exhibited between 44° and 39°:

S-CHOLESTERYL HEXANETHIOATE, melting at 95–96° and clearing at 105°, shows the colors of the visible spectrum at 42–35° on cooling;

S-CHOLESTERYL HEPTANETHIOATE, with a melting and clearing point of 107–109°, exhibits, on cooling, the visible spectrum at 74–73°;

S-CHOLESTERYL OCTANETHIOATE melts and clears at 96–98.5° and shows colors at 75–74° on cooling;

S-CHOLESTERYL EICOSANETHIOATE sinters at 54°, melts at 69°, and clears at 78°. On cooling it becomes cloudy at 77° with a violet color appearing at 49°, and these "crystal colors"[3] remain until the sample has completely crystallized.

INFRARED SPECTRA.[32] The ester carbonyl frequency of S-cholesteryl alkanethioates, observed at 1670–1680 cm^{-1}, is about 50 cm^{-1} lower than that of cholesteryl alkanoates[33] and is in agreement with reported frequencies of thiol esters.[34] The band progression (CH_2-wagging vibrations), observed in cholesteryl alkanoates[33,35] in the region of 1350–1180 cm^{-1} cannot be observed in the alkanethioates. The C-S stretching frequency at 940–950 cm^{-1} is markedly increased over the values found in saturated sulfides (600–700 cm^{-1}).[31,36]

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REFERENCES

1. Elser, W., *Mol. Cryst.* **2**, 1 (1966).
2. Pohlmann, J. L. W., *ibid.* **2**, 15 (1966).
3. Elser, W., *Mol. Cryst. and Liq. Cryst.* **8**, 219 (1969).
4. Elser, W. and Ennulat, R. D., *J. Phys. Chem.* **74**, 1545 (1970).
5. Ennulat, R. D., *Mol. Cryst. and Liq. Cryst.* **8**, 247 (1969).
6. Page, I. H. and Rudy, H., *Biochem. Z.* **220**, 305 (1930).
7. Cataline, E. L., Worrell, L., Jeffries, S. F. and Aronson, S. A., *J. Amer. Pharm. Ass.* **33**, 107 (1944).
8. Gray, G. W., *J. Chem. Soc.* 3733 (1956).
9. Kuksis, A. and Beveridge, J. M. R., *J. Org. Chem.* **25**, 1209 (1960).
10. Mahadevan, V. and Lundberg, W. O., *J. Lipid Res.* **3**, 106 (1962).
11. Staab, H. A., *Angew. Chem.* **71**, 194 (1959).
12. Staab, H. A. and Rohr, W. in "Newer Methods of Preparative Organic Chemistry," W. Foerst, Ed., Verlag Chemie, Weinheim and Academic Press, New York, N.Y., Vol. V, 1968, pp. 74–77.
13. Kaufmann, H. P., Garloff, H. and Deicke, F., *Fette, Seifen, Anstrichm.* **44**, 1037 (1962).
14. Pohlmann, J. L. W. and Elser, W., *Mol. Cryst. and Liq. Cryst.* **8**, 472 (1969).
15. Pohlmann, J. L. W., *ibid.* **8**, 417 (1969).
16. Staab, H. A. and Polenski, B., *Justus Liebigs Ann. Chem.* **655**, 95 (1962).
17. Staab, H. A., Rohr, W. and Mannschreck, A., *Angew. Chem.* **73**, 143 (1961).
18. Staab, H. A. and Mannschreck, A., *Chem. Ber.* **95**, 1284 (1962).
19. Wagner-Jauregg, T. and Lennartz, T., *ibid.* **74**, 27 (1941).
20. King, L. C., Dodson, R. M. and Subluskey, L. A., *J. Amer. Chem. Soc.* **70**, 1176 (1948).
21. Bernstein, S. and Sax, K. J., *J. Org. Chem.* **16**, 685 (1951).
22. Aldrich Chemical Co., Inc., Milwaukee, Wisc.
23. Merck, 0.05–0.2 mm (70–325 mesh).
24. Microanalyses were performed by Schwarzkopf Microanalytical Laboratory, Woodside, N.Y.
25. Ennulat, R. D., *Mol. Cryst.* **3**, 405 (1968).
26. Fieser, L. F., *J. Amer. Chem. Soc.* **75**, 5421 (1953); "Organic Syntheses," Coll. Vol. IV, N. Rabjohn, Ed., J. Wiley & Sons, New York and London, 1963, p. 195.
27. Smith, L. L., Matthews, W. S., Price, J. C., Bachmann, R. C. and Reynolds, B., *J. Chromatog.* **27**, 187 (1967).
28. Metcalfe, L. D. and Schmitz, A. A., *Anal. Chem.* **33**, 363 (1961).
29. Columbia Organic Chemicals, Columbia, South Carolina (Fluka, puriss.).
30. Ennulat, R. D., in "Analytical Calorimetry," R. S. Porter and J. F. Johnson, Ed., Plenum Press, New York, N.Y., 1968, p. 219.
31. Temperatures were taken with a Büchi capillary melting point apparatus and are not corrected.
32. The samples were examined in KBr-disks and the spectra recorded on a Beckman IR-8 spectrophotometer.

33. Labarrère, J. A., Chipault, J. R. and Landberg, W. O., *Anal. Chem.* **30**, 1466 (1958).
34. Baker, A. W. and Harris, G. H., *J. Amer. Chem. Soc.* **82**, 1923 (1960).
35. Carruthers, W. and Plimmer, J. R., *J. Chem. Soc.* 3021 (1962).
36. Meeke, R., Meeke, R. and Littringhaus, A., *Chem. Ber.* **90**, 975 (1957).